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NEWS 21	JAN 27	A new search aid, the Company Name Thesaurus, available in CA/CAplus
NEWS 22	FEB 05	German (DE) application and patent publication number format changes
NEWS 23	MAR 03	MEDLINE and LMEDLINE reloaded
NEWS 24	MAR 03	MEDLINE file segment of TOXCENTER reloaded
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FILE 'HOME' ENTERED AT 13:59:28 ON 24 MAR 2004

=> FIL MEDLINE BIOSIS EMBASE CA SCISEARCH
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FULL ESTIMATED COST	0.21	0.21

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=> s protamine
L1 30366 PROTAMINE

=> s (s (n) protamin?) (n) nls)
UNMATCHED RIGHT PARENTHESIS 'NLS)'
The number of right parentheses in a query must be equal to the number of left parentheses.

=> s (s (n) protamin?) (n) nls))
UNMATCHED RIGHT PARENTHESIS 'NLS))'
The number of right parentheses in a query must be equal to the number of left parentheses.

=> s (s (n) protamin? (n) nls)
L2 4 (S (N) PROTAMIN? (N) NLS)

=> dup rem l2
PROCESSING COMPLETED FOR L2
L3 1 DUP REM L2 (3 DUPLICATES REMOVED)

=> d l3 ibib abs

L3 ANSWER 1 OF 1 MEDLINE on STN DUPLICATE 1
ACCESSION NUMBER: 2002200709 MEDLINE
DOCUMENT NUMBER: PubMed ID: 11906253
TITLE: Protamine-fragment peptides fused to an SV40 nuclear localization signal deliver oligonucleotides that produce antisense effects in prostate and bladder carcinoma cells.
AUTHOR: Benimetskaya Luba; Guzzo-Pernell Nancy; Liu Su-Ting; Lai Johnathan C H; Miller Paul; Stein C A
CORPORATE SOURCE: Howard Florey Institute of Experimental Physiology and Medicine, University of Melbourne, Parkville, Victoria, 3052, Australia.
CONTRACT NUMBER: R01 GM58791 (NIGMS)

SOURCE: Bioconjugate chemistry, (2002 Mar-Apr) 13 (2) 177-87.
Journal code: 9010319. ISSN: 1043-1802.
PUB. COUNTRY: United States
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 200207
ENTRY DATE: Entered STN: 20020406
Last Updated on STN: 20020724
Entered Medline: 20020723

AB The development of antisense technology has focused on improving methods for oligonucleotide delivery into cells. In the present work, we describe a novel strategy for oligonucleotide delivery based on a bifunctional peptide composed of a C-terminal protamine-fragment that contains a DNA-binding domain and an N-terminal nuclear localization signal sequence derived from the SV40 large-T antigen (The sequences of two of the peptides are R6WGR6-PKKKRKV [**s-protamine-NLS**] and R4SR6FGR6VWR4-PKKKRKV [l-protamine-NLS]). We demonstrated, by intrinsic fluorescence quenching, that peptides of this class form complexes with oligodeoxynucleotides. To evaluate delivery, we used a 20-mer phosphorothioate oligomer (Isis 3521) targeted to the 3'-untranslated region of the PKC-alpha mRNA and G3139, an 18-mer phosphorothioate targeted to the first six codons of the human bcl-2 open reading frame, and complexed them with either of two peptides (s- or l-protamine-NLS). These peptides bind to and deliver antisense oligonucleotides to the nucleus of T24 bladder and PC3 prostate cancer cells, as demonstrated by confocal microscopy. Furthermore, as shown by Western and Northern blotting, the peptide-oligonucleotide complexes produced excellent downregulation of the expression of the complementary mRNAs, which in turn resulted in downregulation of protein expression. However, under certain circumstances (predominantly in PC3 cells), incubation of the cells with chloroquine was required to produce antisense activity. Using this strategy, PKC-alpha protein and mRNA expression in T24 and PC3 cells and bcl-2 expression in PC3 cells was reduced by approximately 75 +/- 10% at a minimum concentration of oligomer of 0.25 microM, in combination with 12-15 microM peptide. On the basis of our results, we conclude that arginine-rich peptides of this class may be potentially useful delivery vehicles for the cellular delivery of antisense oligonucleotides. This new strategy may have several advantages over other methods of oligonucleotide delivery and may complement already existing lipid-based technologies.

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=> d his

     (FILE 'HOME' ENTERED AT 13:59:28 ON 24 MAR 2004)

     FILE 'MEDLINE, BIOSIS, EMBASE, CA, SCISEARCH' ENTERED AT 13:59:34 ON 24
     MAR 2004
L1         30366 S PROTAMINE
L2             4 S (S (N) PROTAMIN? (N) NLS)
L3             1 DUP REM L2 (3 DUPLICATES REMOVED)

=> s l1 and (nucle? (2n) local? (2n))
MISSING TERM '2A))'
The search profile that was entered contains a logical operator
followed immediately by a right parenthesis ')'.

=> s l1 and (nucle? (2n) local?)
L4           107 L1 AND (NUCLE? (2N) LOCAL?)

=> dup rem l4
PROCESSING COMPLETED FOR L4
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L5 47 DUP REM L4 (60 DUPLICATES REMOVED)

=> s l5 and py=<2001
2 FILES SEARCHED...
L6 34 L5 AND PY=<2001

=> d l6 1-34 ibib abs

L6 ANSWER 1 OF 34 MEDLINE on STN
ACCESSION NUMBER: 2001324296 MEDLINE
DOCUMENT NUMBER: PubMed ID: 11313824
TITLE: Enhanced cationic liposome-mediated transfection using the DNA-binding peptide mu (mu) from the adenovirus core.
AUTHOR: Murray K D; Etheridge C J; Shah S I; Matthews D A; Russell W; Gurling H M; Miller A D
CORPORATE SOURCE: Department of Psychiatry and Behavioural Sciences, University College London Medical School, Windeyer Institute of Medical Sciences, London, UK.
SOURCE: Gene therapy, **(2001 Mar)** 8 (6) 453-60.
Journal code: 9421525. ISSN: 0969-7128.
PUB. COUNTRY: England: United Kingdom
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 200106
ENTRY DATE: Entered STN: 20010611
Last Updated on STN: 20010611
Entered Medline: 20010607

AB Promising advances in nonviral gene transfer have been made as a result of the production of cationic liposomes formulated with synthetic cationic lipids (cytofectins) that are able to transfect cells. However few cationic liposome systems have been examined for their ability to transfect CNS cells. Building upon our earlier use of cationic liposomes formulated from 3beta-[N-(N',N'-dimethylaminoethane)carbamoyl] cholesterol (DC-Chol) and dioleoyl-L-alpha-phosphatidyl-ethanolamine (DOPE), we describe studies using two cationic viral peptides, mu (mu) and Vp1, as potential enhancers for cationic liposome-mediated transfection. Mu is derived from the condensed core of the adenovirus and was selected to be a powerful nucleic acid charge neutralising and condensing agent. Vp1 derives from the polyomavirus and harbours a classical **nuclear localisation** signal (NLS). Vp1 proved disappointing but lipopolyplex mixtures formulated from pCMVbeta plasmid, mu peptide and DC-Chol/DOPE cationic liposomes were able to transfect an undifferentiated neuronal ND7 cell line with beta-galactosidase reporter gene five-fold more effectively than lipoplex mixtures prepared from pCMVbeta plasmid and DC-Chol/DOPE cationic liposomes. Mu was found to give an identical enhancement to cationic liposome-mediated transfection of ND7 cells as poly-L-lysine (pLL) or **protamine** sulfate (PA). The enhancing effects of mu were found to be even greater (six- to 10-fold) when differentiated ND7 cells were transfected with mu-containing lipopolyplex mixtures. Differentiated ND7 cells represent a simple ex vivo-like post-mitotic CNS cell system. Successful transfection of these cells bodes well for transfection of primary neurons and CNS cells in vivo. These findings have implications for experimental and therapeutic uses of cationic liposome-mediated delivery of nucleic acids to CNS cells.

L6 ANSWER 2 OF 34 MEDLINE on STN
ACCESSION NUMBER: 2000168959 MEDLINE
DOCUMENT NUMBER: PubMed ID: 10706442
TITLE: Gene-delivery systems using cationic polymers.
AUTHOR: Garnett M C
CORPORATE SOURCE: School of Pharmaceutical Sciences, University of Nottingham, University Park, UK..

martin.garnett@nottingham.ac.uk
SOURCE: Critical reviews in therapeutic drug carrier systems, **(1999)** 16 (2) 147-207. Ref: 185
Journal code: 8511159. ISSN: 0743-4863.
PUB. COUNTRY: United States
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
General Review; (REVIEW)
(REVIEW, TUTORIAL)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 200003
ENTRY DATE: Entered STN: 20000327
Last Updated on STN: 20000327
Entered Medline: 20000316

AB Gene therapy will benefit a range of diseases from single-gene defects, to chronic diseases such as cancer, to vaccination. Initially, gene therapy used viral vectors, but the advantages of nonviral systems are now being fully appreciated. This review focuses on cationic polymers as a delivery system for DNA. The physicochemical characterization of DNA polycation complexes that condense and protect DNA from nuclease digestion are considered, together with further factors such as ligand targeting, endosomal escape, and **nuclear localization**. Where possible, the relative efficacy of different cationic polymer delivery systems is compared.

L6 ANSWER 3 OF 34 MEDLINE on STN
ACCESSION NUMBER: 2000165463 MEDLINE
DOCUMENT NUMBER: PubMed ID: 10699729
TITLE: Nuclear gene targeting using negatively charged liposomes.
AUTHOR: Welz C; Neuhuber W; Schreier H; Repp R; Rascher W; Fahr A
CORPORATE SOURCE: Department of Pharmaceutics and Biopharmacy, Philipps University, Ketzerback 63, Marburg, Germany.
SOURCE: International journal of pharmaceutics, **(2000 Mar 10)** 196 (2) 251-2.
Journal code: 7804127. ISSN: 0378-5173.
PUB. COUNTRY: Netherlands
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 200005
ENTRY DATE: Entered STN: 20000518
Last Updated on STN: 20000518
Entered Medline: 20000511

AB Oligonucleotides are a very useful tool to control gene activity. Oligos work by complementary base-pairing with target sequences either in the nucleus or in the cytosol (Zelphati, O., Szoka, F.C., Jr., 1996. Liposomes as a carrier for intracellular delivery of antisense oligonucleotides: a real or magic bullet? J. Contr. Rel. 41, 99-119). In a new approach using chimeric oligonucleotides (Yoon, K., Cole Strauss, A., Kmiec, E.B., 1996. Targeted gene correction of episomal DNA in mammalian cells mediated by a chimeric RNA-DNA oligonucleotide. Proc. Natl. Acad. Sci. USA 93, 2071-2076) conversion of single base mutations with help of intranuclear repair mechanisms maybe an advantageous method to cure genetic diseases which are based on single point mutations. These chimeric oligonucleotides are constructed in a way that they form an intramolecular double strand of DNA and modified RNA-bases. We used a fluorescent labelled pure 68-mer DNA-analogue of a chimeric oligonucleotides to follow the intracellular fate of these kind of genetic material. The oligos were complexed with **protamine** sulfate and coated with three different liposomal formulations. The AVE-3 formulation shows enhanced properties compared to a classical neutral and negatively charged formulation. **Nuclear localisation** of oligos could only be observed with the AVE-3 formulation. Furthermore only the

negatively charged liposome formulations interact with the **protamine**-complexed oligonucleotides.

L6 ANSWER 4 OF 34 MEDLINE on STN
ACCESSION NUMBER: 1999410437 MEDLINE
DOCUMENT NUMBER: PubMed ID: 10480911
TITLE: Ethanol-induced translocation of cAMP-dependent protein kinase to the nucleus. Mechanism and functional consequences.
AUTHOR: Constantinescu A; Diamond I; Gordon A S
CORPORATE SOURCE: Department of Neurology, Ernest Gallo Clinic and Research Center, University of California, San Francisco, California 94110-3518, USA.
CONTRACT NUMBER: RO1 AA10030 (NIAAA)
RO1 AA10039 (NIAAA)
SOURCE: Journal of biological chemistry, **(1999 Sep 17)** 274 (38) 26985-91.
Journal code: 2985121R. ISSN: 0021-9258.
PUB. COUNTRY: United States
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 199910
ENTRY DATE: Entered STN: 19991026
Last Updated on STN: 19991026
Entered Medline: 19991013

AB Ethanol induces translocation of the catalytic subunit (Calpha) of cAMP-dependent protein kinase (PKA) from the Golgi area to the nucleus in NG108-15 cells. Ethanol also induces translocation of the RIIbeta regulatory subunit of PKA to the nucleus; RI and Cbeta are not translocated. Nuclear PKA activity in ethanol-treated cells is no longer regulated by cAMP. Gel filtration and immunoprecipitation analysis confirm that ethanol blocks the reassociation of Calpha with RII but does not induce dissociation of these subunits. Ethanol also reduces inhibition of Calpha by the PKA inhibitor PKI. Pre-incubation of Calpha with ethanol decreases phosphorylation of Leu-Arg-Arg-Ala-Ser-Leu-Gly (Kemptide) and casein but has no effect on the phosphorylation of highly charged molecules such as histone H1 or **protamine**. cAMP-response element-binding protein (CREB) phosphorylation by Calpha is also increased in ethanol-treated cells. This increase in CREB phosphorylation is inhibited by the PKA antagonist (R(p))-cAMPS and by an adenosine receptor antagonist. These results suggest that ethanol affects a cascade of events allowing for sustained **nuclear localization** of Calpha and prolonged CREB phosphorylation. These events may account for ethanol-induced changes in cAMP-dependent gene expression.

L6 ANSWER 5 OF 34 MEDLINE on STN
ACCESSION NUMBER: 1998010146 MEDLINE
DOCUMENT NUMBER: PubMed ID: 9349433
TITLE: **Protamine** sulfate enhances lipid-mediated gene transfer.
AUTHOR: Sorgi F L; Bhattacharya S; Huang L
CORPORATE SOURCE: Department of Pharmacology, University of Pittsburgh School of Medicine, PA 15261, USA.
CONTRACT NUMBER: CA 59327 (NCI)
CA 64654 (NCI)
CA 71731 (NCI)
+
SOURCE: Gene therapy, **(1997 Sep)** 4 (9) 961-8.
Journal code: 9421525. ISSN: 0969-7128.
PUB. COUNTRY: ENGLAND: United Kingdom
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English

FILE SEGMENT: Priority Journals
ENTRY MONTH: 199711
ENTRY DATE: Entered STN: 19971224
Last Updated on STN: 19971224
Entered Medline: 19971120

AB A polycationic peptide, **protamine** sulfate, USP, has been shown to be able to condense plasmid DNA efficiently for delivery into several different types of cells in vitro by several different types of cationic liposomes. The monovalent cationic liposomal formulations (DC-Chol and lipofectin) exhibited increased transfection activities comparable to that seen with the multivalent cationic liposome formulation, lipofectamine. This suggests that lipofectamine's superior in vitro activity arises from its ability to condense DNA efficiently and that **protamine**'s primary role is that of a condensation agent, although it also possesses several amino acid sequences resembling that of a **nuclear localization** signal. While the use of polycations to condense DNA has been previously reported, the of **protamine** sulfate, USP as a condensation agent was found to be superior to poly-L-lysine as well as to various other types of **protamine**. These differences among various salt forms of **protamine** appear to be attributable to structural differences between the **protamines** and not due to differences in the net charge of the molecule. The appearance of lysine residues within the **protamine** molecule correlate with a reduction in binding affinity to plasmid DNA as well as an observed loss in transfection enhancing activity. This finding sheds light on the structural requirements of condensation agents for use in gene transfer protocols. Furthermore, **protamine** sulfate, USP is an FDA-approved compound with a documented safety profile and could be readily used as an adjuvant to a human gene therapy protocol.

L6 ANSWER 6 OF 34 MEDLINE on STN
ACCESSION NUMBER: 96276417 MEDLINE
DOCUMENT NUMBER: PubMed ID: 8694763
TITLE: The variable subunit associated with protein phosphatase 2A0 defines a novel multimember family of regulatory subunits.
AUTHOR: Zolnierowicz S; Van Hoof C; Andjelkovic N; Cron P; Stevens I; Merlevede W; Goris J; Hemmings B A
CORPORATE SOURCE: Friedrich Miescher-Institut, Basel, Switzerland.
SOURCE: Biochemical journal, **(1996 Jul 1)** 317 (Pt 1) 187-94.
Journal code: 2984726R. ISSN: 0264-6021.
PUB. COUNTRY: ENGLAND: United Kingdom
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
OTHER SOURCE: GENBANK-D26445; GENBANK-Z69028; GENBANK-Z69029; GENBANK-Z69030
ENTRY MONTH: 199608
ENTRY DATE: Entered STN: 19960911
Last Updated on STN: 19960911
Entered Medline: 19960823

AB Two protein phosphatase 2A (PP2A) holoenzymes were isolated from rabbit skeletal muscle containing, in addition to the catalytic and PR65 regulatory subunits, proteins of apparent molecular masses of 61 and 56 kDa respectively. Both holoenzymes displayed low basal phosphorylase phosphatase activity, which could be stimulated by **protamine** to an extent similar to that of previously characterized PP2A holoenzymes. Protein micro-sequencing of tryptic peptides derived from the 61 kDa protein, termed PR61, yielded 117 residues of amino acid sequence. Molecular cloning by enrichment of specific mRNAs, followed by reverse transcription-PCR and cDNA library screening, revealed that this protein exists in multiple isoforms encoded by at least three genes, one of which

gives rise to several splicing variants. Comparisons of these sequences with the available databases identified one more human gene and predicted another based on a rabbit cDNA-derived sequence, thus bringing the number of genes encoding PR61 family members to five. Peptide sequences derived from PR61 corresponded to the deduced amino acid sequences of either alpha or beta isoforms, indicating that the purified PP2A preparation was a mixture of at least two trimers. In contrast, the 56 kDa subunit (termed PR56) seems to correspond to the epsilon isoform of PR61. Several regulatory subunits of PP2A belonging to the PR61 family contain consensus sequences for **nuclear localization** and might therefore target PP2A to nuclear substrates.

L6 ANSWER 7 OF 34 MEDLINE on STN
ACCESSION NUMBER: 96192336 MEDLINE
DOCUMENT NUMBER: PubMed ID: 9019163
TITLE: Overexpression of cyclin A in the mammary glands of transgenic mice results in the induction of nuclear abnormalities and increased apoptosis.
AUTHOR: Bortner D M; Rosenberg M P
CORPORATE SOURCE: Glaxo Wellcome, Inc., Research Triangle Park, North Carolina 27709, USA.
SOURCE: Cell growth & differentiation : molecular biology journal of the American Association for Cancer Research, **(1995 Dec)** 6 (12) 1579-89.
Journal code: 9100024. ISSN: 1044-9523.
PUB. COUNTRY: United States
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 199702
ENTRY DATE: Entered STN: 19970227
Last Updated on STN: 19970227
Entered Medline: 19970211

AB Aberrant expression of several cyclin genes has been demonstrated to be associated with many types of tumors. To determine the capacity of cyclin A to function as an oncogene in vivo, wild-type and mutant cyclin A proteins were specifically overexpressed in the mammary glands of transgenic mice using regulatory sequences from the ovine beta-lactoglobulin gene. Several lines of transgenic mice were generated that expressed human cyclin A or a nondegradable mutant version of human cyclin A, in which the amino-terminal 89 amino acids encompassing the cyclin destruction box were removed. The cyclin A transgene products were **localized** in the **nuclei** of mammary epithelial cells, and the transgenic mammary glands had an increase in cyclin A- and cdk2-associated H1 kinase activity. Many mammary epithelial cells in the transgenic glands exhibited nuclear abnormalities, including multinucleation and karyomegaly, which were suggestive of preneoplastic alterations. The abnormalities were more severe in mammary glands of the mutant cyclin A transgenics, which expressed a stabilized cyclin A protein. In situ analysis of mid-lactation mammary gland sections revealed increased numbers of apoptotic cells in the transgenic glands. Double transgenic animals were generated that expressed both the mutant human cyclin A and human cdk2 transgenes, and a more pronounced phenotype resulted. The bigenic mammary glands exhibited focal areas of hyperplasia, as well as a greater incidence of apoptosis than observed in the single transgenic glands, demonstrating in vivo cooperation between these genes in transformation and apoptotic signaling pathways.

L6 ANSWER 8 OF 34 MEDLINE on STN
ACCESSION NUMBER: 95290650 MEDLINE
DOCUMENT NUMBER: PubMed ID: 7772743
TITLE: An immuno-electron microscopic study on the relationship between nuclear matrix and DNA in rat spermatocytes.

AUTHOR: Cheng-Chew S B; Chew E C; Leung P Y; Yang L; Yam H F; Jiao R J; Zhai Z H
CORPORATE SOURCE: Department of Physiology, Chinese University of Hong Kong, Shatin.
SOURCE: In vivo (Athens, Greece), **(1994 Nov-Dec)** 8 (6) 1091-4.
Journal code: 8806809. ISSN: 0258-851X.
PUB. COUNTRY: Greece
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 199507
ENTRY DATE: Entered STN: 19950720
Last Updated on STN: 19950720
Entered Medline: 19950711

AB The nucleus of the mammalian spermatid undergoes a series of changes in its chromatin and nucleoprotein composition during transport from testis to epididymis. The sperm DNA is very tightly packaged by **protamines** instead of histones in somatic cells. However, the nuclear matrix and its association with DNA have not yet been definitively scrutinized with the electron microscope. The present study reveals that the **protamine**-depleted sperm nuclear matrix appears as a network of thick and thin filaments with glodular structures attached the these fibers. Monoclonal antibody to single- and doublestranded DNA was used to localize remnant DNA after extraction. By immunofluorescence microscopy, monoclonal antibody against DNA was **localized** outside the **nucleus** as a halo. Immuno-electron microscopy showed that gold particles were mainly associated with nuclear matrix surrounding the sperm head. Our results suggest a specific structural organization of sperm DNA with its matrix.

L6 ANSWER 9 OF 34 MEDLINE on STN
ACCESSION NUMBER: 94085502 MEDLINE
DOCUMENT NUMBER: PubMed ID: 8262144
TITLE: A temperature-sensitive cell cycle arrest mutation affecting H1 phosphorylation and **nuclear localization** of a small heat shock protein in Tetrahymena thermophila.
AUTHOR: Thatcher T H; Gorovsky M A
CORPORATE SOURCE: Department of Biology, University of Rochester, New York 14627.
CONTRACT NUMBER: GM-26973 (NIGMS)
SOURCE: Experimental cell research, **(1993 Dec)** 209 (2) 261-70.
Journal code: 0373226. ISSN: 0014-4827.
PUB. COUNTRY: United States
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 199401
ENTRY DATE: Entered STN: 19940209
Last Updated on STN: 19940209
Entered Medline: 19940124

AB This report describes a temperature-sensitive Tetrahymena thermophila cell cycle arrest mutant that is also deficient in its heat shock response. Mutants incubated at 41 degrees C undergo rapid dephosphorylation of macronuclear histone H1, in contrast to wild-type cells which hyperphosphorylate H1 under the same conditions. Dephosphorylation is specific to H1 and is associated with a threefold decrease in the level of H1 kinase activity in macronuclei isolated from heat-shocked mutants. A small nuclear heat shock protein, sp29c, is synthesized and phosphorylated normally in the mutant cells but fails to accumulate in macronuclei. Nuclear transport of other heat shock proteins is unaffected. Mutant

cells die slowly at 41 degrees C, a temperature at which wild-type cells resume normal growth after a brief lag. Wild-type cells acquire thermotolerance (competence to survive a 3-h heat shock at 43 degrees C) after a 1-h treatment at 41 degrees C, but mutant cells cannot become thermotolerant and die after the same treatment. The mutation is named chp 1 (cell cycle, heat shock, and phosphorylation defect).

L6 ANSWER 10 OF 34 MEDLINE on STN
ACCESSION NUMBER: 94079722 MEDLINE
DOCUMENT NUMBER: PubMed ID: 8257566
TITLE: The abundant 19-kilodalton protein associated with human sperm nuclei that is related to seminal plasma alpha-inhibins.
AUTHOR: Zalensky A O; Yau P; Breneman J W; Bradbury E M
CORPORATE SOURCE: Department of Biological Chemistry, School of Medicine, University of California at Davis 95616.
SOURCE: Molecular reproduction and development, **(1993 Oct)** 36 (2) 164-73.
Journal code: 8903333. ISSN: 1040-452X.
PUB. COUNTRY: United States
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 199401
ENTRY DATE: Entered STN: 19940203
Last Updated on STN: 19940203
Entered Medline: 19940119

AB A basic protein with a relative molecular mass of 19 kDa has been identified and isolated to purity from sonication-resistant, partially demembranized human sperm nuclei. Several criteria prove that this is the unique sperm-specific protein, which was previously thought to be a sperm/testis histone. Partial primary structure sequencing demonstrates homologies with human seminal alpha-inhibins and semenogelin. From the sequence and Western-blotting data with antibodies against basic seminal inhibin-like peptide, we propose that this 19-kD protein is a product of 52-kDa semenogelin processing. The 19-kDa protein was not found among seminal plasma proteins and may be protected from further cleavage into inhibin-like peptides by its association with the sperm head. Immunofluorescence data indicate its **localization** in the **nuclear** periphery, with preferential concentration at the acrosome calyx boundary.

L6 ANSWER 11 OF 34 MEDLINE on STN
ACCESSION NUMBER: 93244368 MEDLINE
DOCUMENT NUMBER: PubMed ID: 8481469
TITLE: Localization of a spermatid-specific histone 2B protein in mouse spermiogenic cells.
AUTHOR: Moss S B; Orth J M
CORPORATE SOURCE: Department of Obstetrics and Gynecology, University of Pennsylvania School of Medicine, Philadelphia 19104.
CONTRACT NUMBER: HD15563 (NICHD)
HD25524 (NICHD)
SOURCE: Biology of reproduction, **(1993 May)** 48 (5) 1047-56.
Journal code: 0207224. ISSN: 0006-3363.
PUB. COUNTRY: United States
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 199306
ENTRY DATE: Entered STN: 19930618
Last Updated on STN: 19930618
Entered Medline: 19930601

AB Mammalian spermiogenesis is characterized by chromatin condensation and replacement of the histones typical of somatic and earlier spermatogenic cells by **protamines** in the nucleus. However, a spermatid-specific H2b histone (ssH2b) that has an unusual carboxyl-terminus containing a region rich in hydrophobic amino acids is transcribed and translated in mouse round spermatids. The hydrophobicity of this region suggested that the protein may be **localized** at the **nuclear** envelope, the initial site of chromatin condensation during spermiogenesis. To identify ssH2b in the spermatid nucleus, an antiserum (anti-ss-H2b127-138) was generated against a synthetic peptide corresponding to the unique carboxyl-terminus of the protein. Immunocytochemistry of fixed, frozen testicular sections at both the light and electron microscopic levels indicated that ssH2b is present in nuclei of round spermatids. Moreover, the protein was not found to be preferentially associated with the nuclear envelope, indicating that it is not involved in chromatin-nuclear envelope interaction in the round spermatid. Rather, it appeared to be distributed uniformly throughout the nucleus with the exception of its exclusion from the nucleolus. In addition, the ssH2b protein was not found in mature sperm even when the chromatin was partially decondensed, suggesting that it is present and functions only during a restricted period of spermatogenic development.

L6 ANSWER 12 OF 34 MEDLINE on STN
ACCESSION NUMBER: 91347374 MEDLINE
DOCUMENT NUMBER: PubMed ID: 1652372
TITLE: The role of phosphorylation and the CDC28 protein kinase in cell cycle-regulated nuclear import of the S. cerevisiae transcription factor SWI5.
AUTHOR: Moll T; Tebb G; Surana U; Robitsch H; Nasmyth K
CORPORATE SOURCE: Institute for Molecular Pathology, Vienna, Austria.
SOURCE: Cell, **(1991 Aug 23)** 66 (4) 743-58.
Journal code: 0413066. ISSN: 0092-8674.
PUB. COUNTRY: United States
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 199109
ENTRY DATE: Entered STN: 19911020
Last Updated on STN: 20030202
Entered Medline: 19910927

AB The intracellular localization of the S. cerevisiae transcription factor SWI5 is cell cycle dependent. The protein is nuclear in G1 cells but cytoplasmic in S, G2, and M phase cells. We have identified SWI5's **nuclear localization** signal (NLS) and show that it can confer cell cycle-dependent nuclear entry to a heterologous protein. Located within or close to the NLS are three serine residues, mutation of which results in constitutive nuclear entry. These residues are phosphorylated in a cell cycle-dependent manner in vivo, being phosphorylated when SWI5 is in the cytoplasm and dephosphorylated when it is in the nucleus. As all three serines are phosphorylated by purified CDC28-dependent H1 kinase activity in vitro, we propose a model in which the CDC28 kinase acts directly to control nuclear entry of SWI5.

L6 ANSWER 13 OF 34 MEDLINE on STN
ACCESSION NUMBER: 90383813 MEDLINE
DOCUMENT NUMBER: PubMed ID: 2401985
TITLE: Immunocytochemical **localization** of **nuclear protamine** in boar spermatozoa during epididymal transit.
AUTHOR: Rodriguez-Martinez H; Courtens J L; Kvist U; Ploen L
CORPORATE SOURCE: Department of Anatomy and Histology, Faculty of Veterinary Medicine, Swedish University of Agricultural Sciences, Uppsala.

SOURCE: Journal of reproduction and fertility, **(1990 Jul)** 89 (2) 591-5.
Journal code: 0376367. ISSN: 0022-4251.
PUB. COUNTRY: ENGLAND: United Kingdom
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 199010
ENTRY DATE: Entered STN: 19901122
Last Updated on STN: 19901122
Entered Medline: 19901024

AB **Protamine** was specifically demonstrated in boar spermatozoa collected from the rete testis, caput, corpus and cauda epididymidis and the ejaculate by immunoelectron microscopy, using anti-boar or anti-ram **protamine** antisera and an indirect post-embedding immunogold technique. Spermatozoa from all collection sites stained after incubation although with different degrees of labelling. Controls were negative. Labelling increased from the rete testis towards the epididymal corpus, where it was most intense, decreasing sharply thereafter. The weakest binding of the assayed antibodies was obtained in the ejaculated spermatozoa but it could be reversed by in-vitro induction of chromatin decondensation with sodium dodecyl sulphate and the metal-chelating EDTA. The finding of a significant decrease in the immunolabelling detected from the corpus epididymidis onwards indicates a critical point for the interaction between DNA and the **protamines** in boar spermatozoa during the epididymal maturation.

L6 ANSWER 14 OF 34 MEDLINE on STN
ACCESSION NUMBER: 83131918 MEDLINE
DOCUMENT NUMBER: PubMed ID: 6825718
TITLE: Red cell ghost-mediated microinjection of RNA into HeLa cells. II. Cellular translation of **protamine** mRNA; post-translational modifications and nuclear binding of newly-synthesized **protamine**.
AUTHOR: Boogaard C; Dixon G H
SOURCE: Experimental cell research, **(1983 Jan)** 143 (1) 191-205.
Journal code: 0373226. ISSN: 0014-4827.
PUB. COUNTRY: United States
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 198304
ENTRY DATE: Entered STN: 19900318
Last Updated on STN: 19970203
Entered Medline: 19830407

AB Red cell ghosts loaded with **protamine** messenger RNA (pmRNA) were fused to HeLa cells using polyethylene glycol, as a means of introducing the mRNA into heterologous cells. The recipient cells were capable of translating the RNA into the three **protamine** polypeptides, which may be resolved as three peaks (CI, CII, and CIII) by cation exchange chromatography. The synthesis of components CII and CIII was easily observed with possible traces of CI as well. The HeLa cells also phosphorylated CII after synthesis. However, this phosphorylation did not occur with CIII. In addition, CII but not CIII **localized** in the **nucleus** of the HeLa cells after synthesis. Thus, a correlation of post-translational modification with nuclear entry was observed. **Localization** in the **nucleus**, however, was not accompanied by the same tight binding of **protamine** to chromatin as is seen in the homologous trout testis spermatid cells. In the spermatid cells, **protamine** elutes from chromatin at a salt concentration of 1.2 M NaCl. In contrast, in the HeLa cells, the newly synthesized CII which had entered the nucleus, could be eluted with 0.6 M

NaCl. Thus, the tight binding of **protamine** to chromatin in trout testis may require a series of concomitant developmental events, such as core histone hyper-acetylation (Christensen, M E & Dixon, G-H. In press) [17], which would be lacking in the HeLa cells.

L6 ANSWER 15 OF 34 MEDLINE on STN
ACCESSION NUMBER: 83116977 MEDLINE
DOCUMENT NUMBER: PubMed ID: 6185924
TITLE: The localisation of the 5'-termini of in vivo transcripts of a cloned rainbow trout **protamine** gene.
AUTHOR: Gregory S P; Dillon N O; Butterworth P H
SOURCE: Nucleic acids research, **(1982 Dec 11)** 10 (23) 7581-92.
Journal code: 0411011. ISSN: 0305-1048.
PUB. COUNTRY: ENGLAND: United Kingdom
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 198303
ENTRY DATE: Entered STN: 19900318
Last Updated on STN: 19950206
Entered Medline: 19830311

AB The mRNA start site of a cloned rainbow trout **protamine** gene (TPG-3) has been **localised** using S1-**nuclease** mapping and primer extension of in vivo synthesised trout testis poly A+-RNA. The presumptive cap site occurs within an AT-rich region, only 14 nucleotides from the start of the protein-coding sequence. Transcription of this **protamine** gene in vitro, using the Hela whole-cell extract system, generates products initiated at the same nucleotide as that used in vivo. In vitro transcription is abolished by deletion of sequences between -20 and -48, within which is a canonical TATA-box having an 11bp homology with the strong chick conalbumin and Adenovirus-2 major late promoters (CTATAAAAGGG).

L6 ANSWER 16 OF 34 BIOSIS COPYRIGHT 2004 BIOLOGICAL ABSTRACTS INC. on STN
ACCESSION NUMBER: 2001:497834 BIOSIS
DOCUMENT NUMBER: PREV200100497834
TITLE: Utilizing constitutive endocytosis to deliver antisense oligonucleotides into neurons.
AUTHOR(S): Lakkaraju, A. [Reprint author]; Rahman, Y.; Dubinsky, J. M. [Reprint author]
CORPORATE SOURCE: Dept Neurosci, Univ Minnesota, Minneapolis, MN, USA
SOURCE: Society for Neuroscience Abstracts, (2001) Vol. 27, No. 1, pp. 713. print.
Meeting Info.: 31st Annual Meeting of the Society for Neuroscience. San Diego, California, USA. November 10-15, 2001.
ISSN: 0190-5295.
DOCUMENT TYPE: Conference; (Meeting)
Conference; Abstract; (Meeting Abstract)
LANGUAGE: English
ENTRY DATE: Entered STN: 24 Oct 2001
Last Updated on STN: 23 Feb 2002

AB The mechanisms involved in the uptake and intracellular transport of anionic liposomes were investigated in cultured hippocampal neurons. Anionic liposomes encapsulating Cy3-labeled oligonucleotides (Cy3-ON) were internalized in a time- and temperature-dependent manner. Virtually all the neurons imaged exhibited Cy3 fluorescence 30 minutes after incubation with anionic liposomes. Uptake occurred by receptor-mediated endocytosis via LRP, the LDL receptor-related protein, independent of cell surface proteoglycans, as demonstrated by sensitivity to hyperosmolar sucrose, the LRP receptor antagonist RAP, and heparin and **protamine** sulfate. Liposomal endocytosis required clathrin, dynamin, an intact cytoskeletal

network and phosphatidylinositol 3-kinase activity, as demonstrated by sensitivity to the calcineurin inhibitor FK506, the PI3 kinase inhibitor wortmannin, and the microtubule and actin depolymerizers nocodozole and cytochalasin B. **Nuclear localization** of Cy3-ON was observed within 1-3 h of incubation, suggesting that this delivery system bypassed the lysosomal pathway and the oligonucleotides were freely available to the neuron. Maximally 25% of Cy3-ON were colocalized with Oregon Green-transferin indicating that ON did not recyle rapidly. Liposomal lipids containing N-Rh-DOPE were recycled back to the cell surface via transferrin-containing compartments. Uptake of cationic lipid-oligonucleotide complexes and oligonucleotides without lipid vectors was much lower than that of oligonucleotides delivered by anionic liposomes. Anionic liposomes can be exploited to deliver a variety of substances to the cell interior.

L6 ANSWER 17 OF 34 BIOSIS COPYRIGHT 2004 BIOLOGICAL ABSTRACTS INC. on STN
ACCESSION NUMBER: 1997:273895 BIOSIS
DOCUMENT NUMBER: PREV199799565613
TITLE: Histones, **protamine**, and polylysine but not poly(E:K) enhance transfection efficiency.
AUTHOR(S): Boulikas, Teni [Reprint author]; Martin, Francis
CORPORATE SOURCE: Inst. Molecular Med. Sci., 460 Page Mill Road, Palo Alto, CA 94306, USA
SOURCE: International Journal of Oncology, (1997) Vol. 10, No. 2, pp. 317-322.
ISSN: 1019-6439.
DOCUMENT TYPE: Article
LANGUAGE: English
ENTRY DATE: Entered STN: 24 Jun 1997
Last Updated on STN: 24 Jun 1997

AB Liposomal gene delivery has a great potential for the treatment of cancer and other human diseases. In this work we have investigated the optimal conditions for liposome-mediated transfer of the luciferase gene to human erythroleukemia K562 cells. DDAB:DOPE liposomes were more efficient than lysyl-DOPE:DOPE (1:2) and DDAB:cholesterol for transfection. Total histones from bovine thymus, salmon sperm **protamine**, and polylysine at an optimal ratio of 0.5 mg protein/mg DNA enhanced up to 7-fold the transfection efficiency of luciferase plasmids; on the contrary, the synthetic polymer poly (E:K), containing glutamic acid and lysine residues in a random order at a ratio of 1:4, diminished luciferase expression. Transfection was nearly zero at high histone:DNA ratio by reversal of the charge of the particle from negative to positive leading to its inability to interact with cationic liposomes. The increase in luciferase gene expression by DNA-binding proteins might arise from an increased transfer across the cell membrane of the liposome-DNA-protein complex but also by an increase in nuclear import of the DNA-protein complex because of the presence of **nuclear localization** signals on the protein molecule used for DNA condensation.

L6 ANSWER 18 OF 34 BIOSIS COPYRIGHT 2004 BIOLOGICAL ABSTRACTS INC. on STN
ACCESSION NUMBER: 1977:235735 BIOSIS
DOCUMENT NUMBER: PREV197764058099; BA64:58099
TITLE: ESTROGEN RECEPTOR UNOCCUPIED SITES IN NUCLEI OF A BREAST TUMOR CELL LINE.
AUTHOR(S): ZAVA D T; MCGUIRE W L
SOURCE: Journal of Biological Chemistry, (1977) Vol. 252, No. 11, pp. 3703-3708.
CODEN: JBCHA3. ISSN: 0021-9258.
DOCUMENT TYPE: Article
FILE SEGMENT: BA
LANGUAGE: Unavailable

AB Normal estrogen responsive tissues such as the rat uterus contain unoccupied cytoplasmic receptors (Rc) for estrogen (E). After binding

estradiol, these receptors are translocated into the nucleus. In contrast to normal target tissues, nuclei of the human breast cancer cell line MCF-7 contain high levels of estrogen receptor (Rn) even when grown in estrogen-free serum. This **nuclear localization** of Rn is demonstrated by a **protamine** sulfate assay. Rn is extracted in 0.6 M KCl, quantitatively precipitated with **protamine** sulfate and incubated with [3H]estradiol. Incubation at 4° C (15 h) and 37° C (2.5 h) measures unoccupied (Rn) and total (Rn + RnE) receptor, respectively. Cytosol from confluent cells grown in the absence of estradiol contains only 0.5 pmol of free Rc/mg of DNA, while a nuclear KCl extract contains 1.5 pmol of free Rn/mg of DNA. Rc and Rn have high affinity binding for estradiol .**GRAPHIC**. = 0.3 mM, .**GRAPHIC**. = 0.8 nM, 4° C) and sediment in 0.4 M KCl sucrose gradients at 4-5 S. Low salt buffers result in a 9 S component for Rc, whereas Rn remains in the 4-5 S region. When purified intact nuclei were incubated directly with increasing concentrations of [3H]estradiol, similar levels of Rn (1.2 pmol/mg of DNA) and the same affinity for the ligand were found, thus demonstrating the same **nuclear localization** of estrogen receptor by other techniques. Receptors for progesterone and androgens are found in the cytosol (90% or more). In rapidly growing cells, Rc and Rn are significantly higher than in confluent cells (0.8 pmol/mg of DNA and 5 pmol/mg of DNA, respectively). At least 1 other breast cancer cell line which contains high levels of RN was found.

L6 ANSWER 19 OF 34 EMBASE
on STN

ACCESSION NUMBER: 2000368573 EMBASE
TITLE: Roughex mediates G1 arrest through a physical association with cyclin A.
AUTHOR: Avedisov S.N.; Krasnoselskaya I.; Mortin M.; Thomas B.J.
CORPORATE SOURCE: B.J. Thomas, National Cancer Institute, NIH, Bldg. 37, 37 Convent Dr., Bethesda, MD 20892-4255, United States. bthomas@sunspot.nci.nih.gov
SOURCE: Molecular and Cellular Biology, (2000) 20/21 (8220-8229). Refs: 44
ISSN: 0270-7306 CODEN: MCEBD4
COUNTRY: United States
DOCUMENT TYPE: Journal; Article
FILE SEGMENT: 021 Developmental Biology and Teratology
029 Clinical Biochemistry
LANGUAGE: English
SUMMARY LANGUAGE: English
AB Differentiation in the developing Drosophila eye requires synchronization of cells in the G1 phase of the cell cycle. The roughex gene product plays a key role in this synchronization by negatively regulating cyclin A protein levels in G1. We show here that coexpressed Roughex and cyclin A physically interact in vivo. Roughex is a nuclear protein, while cyclin A was previously shown to be exclusively cytoplasmic during interphase in the embryo. In contrast, we demonstrate that in interphase cells in the eye imaginal disk cyclin A is present in both the nucleus and the cytoplasm. In the presence of ectopic Roughex, cyclin A becomes strictly nuclear and is later degraded. Nuclear targeting of both Roughex and cyclin A under these conditions is dependent on a C-terminal **nuclear localization** signal in Roughex. Disruption of this signal results in cytoplasmic localization of both Roughex and cyclin A, confirming a physical interaction between these molecules. Cyclin A interacts with both Cdc2 and Cdc2c, the Drosophila Cdk2 homolog, and Roughex inhibits the histone H1 kinase activities of both cyclin A-Cdc2 and cyclin A-Cdc2c complexes in whole-cell extracts. Two-hybrid experiments suggested that the inhibition of kinase activity by Roughex results from competition with the cyclin-dependent kinase subunit for binding to cyclin A. These findings suggest that Roughex can influence the intracellular distribution of cyclin A and define Roughex as a distinct

and specialized cell cycle inhibitor for cyclin A-dependent kinase activity.

L6 ANSWER 20 OF 34 EMBASE COPYRIGHT 2004 ELSEVIER INC. ALL RIGHTS RESERVED. on STN

ACCESSION NUMBER: 1999288882 EMBASE
TITLE: A maternal form of the phosphatase Cdc25A regulates early embryonic cell cycles in Xenopus laevis.
AUTHOR: Kim S.H.; Li C.; Maller J.L.
CORPORATE SOURCE: J.L. Maller, Program in Molecular Biology, Department of Pharmacology, Univ. of Colorado School of Medicine, Denver, CO 80262, United States. Jim.Maller@uchsc.edu
SOURCE: Developmental Biology, (15 Aug 1999) 212/2 (381-391). Refs: 47
ISSN: 0012-1606 CODEN: DEBIAO
COUNTRY: United States
DOCUMENT TYPE: Journal; Article
FILE SEGMENT: 021 Developmental Biology and Teratology
LANGUAGE: English
SUMMARY LANGUAGE: English

AB In mammalian cells the Cdc25 family of dual-specificity phosphatases has three distinct isoforms, termed A, B, and C, which are thought to play discrete roles in cell-cycle control. In this paper we report the cloning of Xenopus Cdc25A and demonstrate its developmental regulation and key role in embryonic cell-cycle control. Northern and Western blot analyses show that Cdc25A is absent in oocytes, and synthesis begins within 30 min after fertilization. The protein product is **localized** in the **nucleus** in interphase and accumulates continuously until the midblastula transition (MBT), after which it is degraded. Upon injection into newly fertilized eggs, wild-type Cdc25A shortened the cell cycle and accelerated the timing of cleavage, whereas embryos injected with phosphatase-dead Cdc25A displayed a dose- dependent increase in the length of the cell cycle and a slower rate of cleavage. In contrast, injection of the phosphatase-dead Cdc25C isoform had no effect. Western blotting with an antibody specific for phosphorylated tyr15 in Cdc2/Cdk2 revealed a cycle of phosphorylation/dephosphorylation in each cell cycle in control embryos, and in embryos injected with phosphatase- dead Cdc25A there was a twofold increase in the level of p-tyr in Cdc2/Cdk2. Consistent with this, the levels of cyclin B/Cdc2 and cyclin E/Cdk2 histone H1 kinase activity were both reduced by approximately 50% after phosphatase- dead Cdc25A injection. The phosphatase-dead Cdc25A could be recovered in a complex with both Cdks, suggesting that it acts in a dominant-negative fashion. These results indicate that periodic phosphorylation of Cdc2/Cdk2 on tyr15 occurs in each pre-MBT cell cycle, and dephosphorylation of Cdc2/Cdk2 by Cdc25A controls at least in part the length of the cell cycle and the timing of cleavage in pre-MBT embryos. The disappearance of Cdc25A after the MBT may underlie in part the lengthening of the cell cycle at that time.

L6 ANSWER 21 OF 34 EMBASE COPYRIGHT 2004 ELSEVIER INC. ALL RIGHTS RESERVED. on STN

ACCESSION NUMBER: 97036964 EMBASE
DOCUMENT NUMBER: 1997036964
TITLE: **Nuclear localization** of cyclin B1 mediates its biological activity and is regulated by phosphorylation.
AUTHOR: Li J.; Meyer A.N.; Donoghue D.J.
CORPORATE SOURCE: D.J. Donoghue, Department of Chemistry/Biochemistry, Center for Molecular Genetics, University of California, San Diego, CA 92093-0367, United States. ddonoghue@ucsd.edu
SOURCE: Proceedings of the National Academy of Sciences of the United States of America, (1997) 94/2 (502-507). Refs: 50

ISSN: 0027-8424 CODEN: PNASA6
COUNTRY: United States
DOCUMENT TYPE: Journal; Article
FILE SEGMENT: 029 Clinical Biochemistry
LANGUAGE: English
SUMMARY LANGUAGE: English

AB M-phase promoting factor or maturation promoting factor, a key regulator of the G2 → M transition of the cell cycle, is a complex of cdc2 and a B- type cyclin. We have previously shown that Xenopus cyclin B1 has five sites of Ser phosphorylation, four of which map to a recently identified cytoplasmic retention signal (CRS). The CRS appears to be responsible for the cytoplasmic localization of B-type cyclins, although the underlying mechanism is still unclear. Phosphorylation of cyclin B1 is not required for cdc2 binding or cdc2 kinase activity. However, when all of the Ser phosphorylation sites in the CRS are mutated to Ala to abolish phosphorylation, the mutant cyclin B1(Ala) is inactivated; activity can be enhanced by mutation of these residues to Glu to mimic phosphoserine, suggesting that phosphorylation of cyclin B1 is required for its biological activity. Here we show that biological activity can be restored to cyclin B1(Ala) by appending either a **nuclear localization** signal (NLS), or a second CRS domain with the Ser phosphorylation sites mutated to Glu, while fusion of a second CRS domain with the Ser phosphorylation sites mutated to Ala inactivates wild-type cyclin B1. Nuclear histone H1 kinase activity was detected in association with cyclin B1(Ala) targeted to the nucleus by a wild-type NLS, but not by a mutant NLS. These results demonstrate that nuclear translocation mediates the biological activity of cyclin B1 and suggest that phosphorylation within the CRS domain of cyclin B1 plays a regulatory role in this process. Furthermore, given the similar in vitro substrate specificity of cyclin-dependent kinases, this investigation provides direct evidence for the hypothesis that the control of subcellular localization of cyclins plays a key role in regulating the biological activity of cyclin-dependent kinase-cyclin complexes.

L6 ANSWER 22 OF 34 EMBASE on STN
ACCESSION NUMBER: 95121468 EMBASE
DOCUMENT NUMBER: 1995121468
TITLE: Cloning of p57(KIP2), a cyclin-dependent kinase inhibitor with unique domain structure and tissue distribution.
AUTHOR: Lee M.-H.; Reynisdottir I.; Massague J.
CORPORATE SOURCE: Cell Biology and Genetics Program, Howard Hughes Medical Institute, Memorial Sloan-Kettering Cancer Ctr.,New York, NY 10021, United States
SOURCE: Genes and Development, (1995) 9/6 (639-649).
ISSN: 0890-9369 CODEN: GEDEEP
COUNTRY: United States
DOCUMENT TYPE: Journal; Article
FILE SEGMENT: 022 Human Genetics
029 Clinical Biochemistry
LANGUAGE: English
SUMMARY LANGUAGE: English

AB Progression through the cell cycle is catalyzed by cyclin-dependent kinases (CDKs) and is negatively controlled by CDK inhibitors (CDIs). We have isolated a new member of the p21(CIP1)/p27(KIP1) CDI family and named it p57(KIP2) to denote its apparent molecular mass and higher similarity to p27(KIP1). Three distinct p57 cDNAs were cloned that differ at the start of their open reading frames and correspond to messages generated by the use of distinct splice acceptor sites. p57 is distinguished from p21 and p27 by its unique domain structure. Four distinct domains follow the heterogeneous amino-terminal region and include, in order, a p21/p27-related CDK inhibitory domain, a proline-rich (28% proline) domain, an acidic (36% glutamic or aspartic acid) domain, and a

carboxy-terminal nuclear targeting domain that contains a putative CDK phosphorylation site and has sequence similarity to p27 but not to p21. Most of the acidic domain consists of a novel, tandemly repeated 4-amino acid motif. p57 is a potent inhibitor of G1- and S-phase CDKs (cyclin E-cdk2, cyclin D2-cdk4, and cyclin A-cdk2) and, to lesser extent, of the mitotic cyclin B-Cdc2. In mammalian cells, p57 **localizes** to the **nucleus**, associates with G1 CDK components, and its overexpression causes a complete cell cycle arrest in G1 phase. In contrast to the widespread expression of p21 and p27 in human tissues, p57 is expressed in a tissue-specific manner, as a 1.5-kb species in placenta and at lower levels in various other tissues and a 7-kb mRNA species observed in skeletal muscle and heart. The expression pattern and unique domain structure of p57 suggest that this CDI may play a specialized role in cell cycle control.

L6 ANSWER 23 OF 34 EMBASE COPYRIGHT 2004 ELSEVIER INC. ALL RIGHTS RESERVED. on STN

ACCESSION NUMBER: 93021342 EMBASE
DOCUMENT NUMBER: 1993021342
TITLE: Adenovirus DNA polymerase is a phosphoprotein.
AUTHOR: Ramachandra M.; Nakano R.; Mohan P.M.; Rawitch A.B.; Padmanabhan R.
CORPORATE SOURCE: Biochemistry/Molecular Biology Dept., University of Kansas Medical Center, 3901 Rainbow Blvd.,Kansas City, KS 66160-7421, United States
SOURCE: Journal of Biological Chemistry, (1993) 268/1 (442-448). ISSN: 0021-9258 CODEN: JBCHA3
COUNTRY: United States
DOCUMENT TYPE: Journal; Article
FILE SEGMENT: 004 Microbiology
LANGUAGE: English
SUMMARY LANGUAGE: English

AB Biological activities of many of the eukaryotic DNA replication proteins are modulated by protein phosphorylation. Investigations of the phosphorylation of adenovirus DNA polymerase (AdPol) have been difficult mainly because of its low level of synthesis in adenovirus-infected HeLa cells. However, when AdPol was overproduced using the recombinant vaccinia virus (RV-AdPol) and the baculovirus expression systems, or by a large scale metabolic labeling of adenovirus 2-infected HeLa cells (native AdPol), in vivo phosphorylation of AdPol could be demonstrated. Phosphoamino acid analysis of [32P]AdPol indicated the presence of phosphoserine independent of the source of AdPol. Comparison of tryptic peptide maps of native AdPol and RV-AdPol revealed that the majority of phosphopeptides were common. Fractionation by high performance liquid chromatography and sequencing of one of the major phosphopeptides revealed serine 67 as a site of phosphorylation. Interestingly, this site is located close to the **nuclear localization** signal of AdPol and has a consensus substrate recognition sequence for histone H1 (cdc2-related) kinases and mitogen-activated protein kinases. Dephosphorylation of AdPol with calf intestinal alkaline phosphatase resulted in significant decrease in its activity in the in vitro DNA replication initiation assay, suggesting that phosphorylation is important for its biological activity.

L6 ANSWER 24 OF 34 EMBASE COPYRIGHT 2004 ELSEVIER INC. ALL RIGHTS RESERVED. on STN

ACCESSION NUMBER: 92268607 EMBASE
DOCUMENT NUMBER: 1992268607
TITLE: Expression of homeobox genes during spermatogenesis.
AUTHOR: Wolgemuth D.J.; Viviano C.M.; Watrin F.
CORPORATE SOURCE: Department of Genetics/Development, Center for Reproductive Sciences, Columbia Univ. Phys./Surgeons Coll., 630 West 168th Street,New York, NY 10032, United States

SOURCE: Annals of the New York Academy of Sciences, (1991) 637/-(300-312).
ISSN: 0077-8923 CODEN: ANYAA
COUNTRY: United States
DOCUMENT TYPE: Journal; Conference Article
FILE SEGMENT: 021 Developmental Biology and Teratology
029 Clinical Biochemistry
LANGUAGE: English
SUMMARY LANGUAGE: English

AB The discovery of the evolutionarily conserved homeobox sequence provided a means for isolating potential development regulating genes in vertebrates, and indeed, homeobox-containing genes have now been isolated from a variety of species from Drosophila to human. Over thirty homeobox-containing genes have been isolated from mouse alone, and like their Drosophila counterparts, they are thought to play an important role in pattern formation during embryonic development. In addition, they appear to be important in the differentiation of several adult tissues including the nervous system, the hematopoietic system, and the germ cell lineage. Conserved features of Hox proteins, their **nuclear localization**, and their ability to regulate the expression of other genes has suggested that these proteins function as transcription factors. The generation of transgenic mice in which germ cell specific expression of Hox-1.4 has been altered while somatic expression remains intact would provide a means of assessing the action of Hox-1.4 during germ cell development. Once testis-specific promoter elements have been defined for Hox-1.4, they could be targeted for mutation by homologous recombination. Alternatively, Hox-1.4 antisense constructs could be designed such that they would be driven by an appropriate spermatogenic-specific promoter (for example PGK-2) and they would produce stable RNAs. Stabilization of the antisense RNAs might be accomplished using sequences from the 3' untranslated region of the **protamine** -1 gene. The antisense experiments are more immediately feasible as the testis specific promoter elements for Hox-1.4 have not yet been defined. It is important however, that efforts be made to interfere with the expression of this gene as these types of experiments are likely to provide valuable information regarding function during spermatogenesis.

L6 ANSWER 25 OF 34 EMBASE COPYRIGHT 2004 ELSEVIER INC. ALL RIGHTS RESERVED. on STN
ACCESSION NUMBER: 75111199 EMBASE
DOCUMENT NUMBER: 1975111199
TITLE: Effects of basic proteins and heparin on early development of Xenopus laevis (Hungarian).
AUTHOR: Do N.K.; Nagy U.; Csaba G.
CORPORATE SOURCE: Biol. Int., Semmelweis Orvostud. Egyet., Budapest, Hungary
SOURCE: BIOL.KOZL., (1973) 21/1-2 (55-59).
CODEN: BIKOAP
DOCUMENT TYPE: Journal
FILE SEGMENT: 037 Drug Literature Index
021 Developmental Biology and Teratology
030 Pharmacology
LANGUAGE: Hungarian

AB Neither exogenous **protamine** nor polylysin had any significant effect on the morphogenesis of Xenopus embryos, however the embryos were destroyed and their growth retarded. With the early administration of heparin the embryos died and with its late administration pigmentation increased. The tritium labeled **protamine** sulfate penetrates into the micromeres and can be **localized** in the **nucleus** as well as in the cytoplasm.

L6 ANSWER 26 OF 34 CA COPYRIGHT 2004 ACS on STN
ACCESSION NUMBER: 136:212638 CA
TITLE: Identification and characterization of CaMKP-N,

nuclear calmodulin-dependent protein kinase phosphatase

AUTHOR(S): Takeuchi, Masayuki; Ishida, Atsuhiko; Kameshita, Isamu; Kitani, Takako; Okuno, Sachiko; Fujisawa, Hitoshi

CORPORATE SOURCE: Department of Biochemistry, Asahikawa Medical College, Asahikawa, 078-8510, Japan

SOURCE: Journal of Biochemistry (Tokyo, Japan) (**2001**), 130(6), 833-840
CODEN: JOBIAO; ISSN: 0021-924X

PUBLISHER: Japanese Biochemical Society

DOCUMENT TYPE: Journal

LANGUAGE: English

AB Calmodulin-dependent protein kinase phosphatase (CaMKP) dephosphorylates and concomitantly deactivates multifunctional Ca2+/calmodulin-dependent protein kinases (CaMKs), such as CaMKI, CaMKII, and CaMKIV. In the present study, a nuclear CaMKP-related protein, CaMKP-N, was identified. This protein consisted of 757 amino acid residues with a calculated mol. weight of 84,176. Recombinant CaMKP-N dephosphorylated CaMKIV. The activity of CaMKP-N requires Mn2+ ions and is stimulated by polycations. Transiently expressed CaMKP-N in COS-7 cells was **localized** in the **nucleus**. This finding together with previous reports regarding localization of CaMKs indicates that CaMKP-N dephosphorylates CaMKIV and nuclear CaMKII, whereas CaMKP dephosphorylates CaMKI and cytosolic CaMKII.

REFERENCE COUNT: 37 THERE ARE 37 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L6 ANSWER 27 OF 34 CA COPYRIGHT 2004 ACS on STN

ACCESSION NUMBER: 136:195999 CA

TITLE: Identification of a specific sperm nuclei selenoenzyme necessary for **protamine** thiol cross-linking during sperm maturation

AUTHOR(S): Pfeifer, Henning; Conrad, Marcus; Roethlein, Doris; Kyriakopoulos, Antonios; Brielmeier, Markus; Bomkamm, Georg W.; Behne, Dietrich

CORPORATE SOURCE: Department Trace Elements in Health and Nutrition, Hahn-Meitner-Institut Berlin, Berlin, D-14109, Germany

SOURCE: FASEB Journal (**2001**), 15(7), 1236-1238, 10.1096/fj.00-0655fje
CODEN: FAJOEC; ISSN: 0892-6638

PUBLISHER: Federation of American Societies for Experimental Biology

DOCUMENT TYPE: Journal

LANGUAGE: English

AB A 34 kD selenoprotein purified from rat testis was identified as a specific sperm nuclei glutathione peroxidase (snGPx) with similar properties to phospholipid hydroperoxide glutathione peroxidase (PHGPx). The determination of its primary structure by anal. of its first N-terminal amino acids, database search, polymerase chain reaction, and sequencing of the cDNA showed that it differs from PHGPx in its N-terminal sequence. This sequence, which is encoded for by an alternative exon in the first intron of the PHGPx gene, shows more than 50% homol. to the **protamine** sequences and contains a **nuclear localization** signal. In rats, snGPx is highly expressed in the nuclei of the late spermatids where it is the only selenoprotein present. Its appearance coincides with the reorganization of DNA, which leads to highly condensed chromatin stabilized by cross-linked **protamine** thiols. In selenium-depleted rats where the concentration of snGPx had decreased to one-third of the normal level, chromatin condensation was severely disturbed. We provided evidence that snGPx acts as a **protamine** thiol peroxidase responsible for disulfide crosslinking by reduction of reactive oxygen species. Its dual function in chromatin condensation and

the protection of sperm DNA against oxidation is necessary to ensure male fertility and sperm quality.

REFERENCE COUNT: 28 THERE ARE 28 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L6 ANSWER 28 OF 34 CA COPYRIGHT 2004 ACS on STN
ACCESSION NUMBER: 134:338733 CA
TITLE: Controlling sex ratios in animal breeding by sex-chromosome selective control of sperm viability
INVENTOR(S): Mileham, Alan; Affara, Nabeel; Plastow, Graham
PATENT ASSIGNEE(S): PIG Improvement Co. (Uk) Ltd., UK
SOURCE: PCT Int. Appl., 25 pp.
CODEN: PIXXD2
DOCUMENT TYPE: Patent
LANGUAGE: English
FAMILY ACC. NUM. COUNT: 1
PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
WO 2001032008	A1	20010510	WO 2000-GB4223	20001103 <--
W: AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, BZ, CA, CH, CN, CR, CU, CZ, DE, DK, DM, DZ, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NO, NZ, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TR, TT, TZ, UA, UG, US, UZ, VN, YU, ZA, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM				
RW: GH, GM, KE, LS, MW, MZ, SD, SL, SZ, TZ, UG, ZW, AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, TR, BF, BJ, CF, CG, CI, CM, GA, GN, GW, ML, MR, NE, SN, TD, TG				
BR 2000015313	A	20020702	BR 2000-15313	20001103
EP 1272030	A1	20030108	EP 2000-973035	20001103
R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, LU, NL, SE, MC, PT, IE, SI, LT, LV, FI, RO, MK, CY, AL, TR				
US 2003087860	A1	20030508	US 2002-137706	20020503

PRIORITY APPLN. INFO.: GB 1999-26161 A 19991104
WO 2000-GB4223 W 20001103

AB Methods for the control of sex ratio in non-human mammals are provided. These methods involve the production of transgenic animals which have particular transgenes integrated into their genomes. Animals produced using such methods are also provided, as are the transgene constructs. The method can be used to selectively inhibit function of either X or Y chromosome by placing a transgene that will inhibit sperm function under control of a promoter that only functions in post-meiotic spermatids.

REFERENCE COUNT: 5 THERE ARE 5 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L6 ANSWER 29 OF 34 CA COPYRIGHT 2004 ACS on STN
ACCESSION NUMBER: 129:118754 CA
TITLE: Method for making a compound for delivery to cells by forming a polymer in the presence of a template drug, especially nucleic acid
INVENTOR(S): Wolff, Jon A.; Hagstrom, James E.; Budker, Vladimir G.; Trubetskoy, Vladimer S.; Slattum, Paul M.; Hanson, Lisa J.
PATENT ASSIGNEE(S): Mirus Corp., USA
SOURCE: PCT Int. Appl., 79 pp.
CODEN: PIXXD2
DOCUMENT TYPE: Patent
LANGUAGE: English
FAMILY ACC. NUM. COUNT: 6
PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
WO 9829541	A1	19980709	WO 1997-US24089	19971230 <--
RW: AT, BE, CH, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE				
US 6126964	A	20001003	US 1997-778657	19970103 <--
EP 958356	A1	19991124	EP 1997-954803	19971230 <--
R: AT, BE, CH, DE, DK, ES, FR, GB, IT, LI, NL, SE, IE				
US 2002061287	A1	20020523	US 2001-4763	20011205
US 2002085989	A1	20020704	US 2001-5294	20011205

PRIORITY APPLN. INFO.: US 1997-778657 A 19970103
US 1996-9593P P 19960104
WO 1997-US24089 W 19971230
US 1999-464871 A3 19991216

OTHER SOURCE(S): MARPAT 129:118754

AB A method of making a compound for delivery to a cell comprising forming a polymer in the presence of a biol. active drug is disclosed.. A method of forming polymers in the presence of nucleic acid using template polymerization and of having the polymerization occur in heterophase systems is further disclosed. These methods can be used for the delivery of nucleic acids, for condensing the nucleic acid, for forming nucleic acid-binding polymers, for forming supramol. complexes containing nucleic acid and polymer, and for forming an interpolyelectrolyte complex. The **nuclear localizing** peptide of SV40 T antigen was copolymd. with dithiobis[succinimidylpropionate] in the presence of plasmid DNA and this process enabled the formation of complexes that expressed luciferase after transfection into 3T3 cells in culture.

REFERENCE COUNT: 4 THERE ARE 4 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L6 ANSWER 30 OF 34 CA COPYRIGHT 2004 ACS on STN

ACCESSION NUMBER: 125:318049 CA

TITLE: Perinuclear localization of an intracellular binding protein related to the fibroblast growth factor (FGF) receptor 1 is temporally associated with the nuclear trafficking of FGF-2 in proliferating epiphyseal growth plate chondrocytes

AUTHOR(S): Kilkenny, Dawn M.; Hill, David J.

CORPORATE SOURCE: Lawson Research Institute, St. Joseph's Health Centre, London, ON, N6A 4V2, Can.

SOURCE: Endocrinology (**1996**), 137(11), 5078-5089
CODEN: ENDOAO; ISSN: 0013-7227

PUBLISHER: Endocrine Society

DOCUMENT TYPE: Journal

LANGUAGE: English

AB Fibroblast growth factor-2 (FGF-2) is a potent autocrine mitogen for fetal epiphyseal growth plate chondrocytes and exhibits a transient nuclear translocation during G1 of the cell cycle. The authors have characterized an intracellular binding protein (FGFBP) for FGF-2 that undergoes a juxtanuclear localization coincident with the nuclear translocation of the growth factor. Chondrocytes were isolated from the proliferative zone of the ovine fetal proximal tibial growth plate at 50-130 days gestation by collagenase digestion and were maintained in monolayer at earlier passage number Cells were growth restricted by serum starvation for 48 h, and the synchronized culture was restarted into the cell cycle in the presence of 2% FBS. Cells were removed between 4-26 h of incubation,and fractions representing the plasma membrane, cytoplasm, nuclear membrane, and nuclear contents were separated by differential centrifugation. FGFBPs were separated using FGF-2 affinity chromatog. Ligand blot anal. using 125I-labeled FGF-2 showed that a FGFBP of 46-48 kDa (represented by a double band) was present on the nuclear membrane at mid to late G1, and Western blot showed this to be immunol. related to a part of the extracellular domain of the high affinity FGF receptor 1 (FGFR1). Immunocytochem. with intact cell cultures showed that this protein underwent a juxtanuclear distribution

through mid to late G1. Immunopptn. was performed to monitor newly synthesized FGFR1 migration throughout the cell cycle. Synchronized cells were cultured in medium containing 35S-labeled methionine/cysteine, and the cellular compartments were separated before immunopptn. using an antibody raised against the extracellular domain of FGFR1. Newly synthesized FGFR1-related proteins appeared throughout G1 and migrated multi-directionally within the cell; intact receptor of 125-145 kDa accumulated at the plasma membrane, while both intact receptor and truncated FGFR1 of 46-48 kDa were detected on the nuclear membrane, but not within the nucleus. Cells were incubated with **protamine** sulfate to prevent the binding of endogenous, cell membrane-associated FGF-2 to high affinity FGFRs and their subsequent internalization. This did not alter the juxtanuclear accumulation of truncated FGFR1 in late G1, suggesting that this was not derived from the plasma membrane. The truncated FGFR1 may mediate the nuclear translocation of FGF-2 during late G1.

L6 ANSWER 31 OF 34 CA COPYRIGHT 2004 ACS on STN
ACCESSION NUMBER: 120:188220 CA
TITLE: Poly(A)+ ribonucleic acids are enriched in spermatocyte nuclei but not in chromatoid bodies in the rat testis
AUTHOR(S): Morales, Carlos R.; Hecht, Normal B.
CORPORATE SOURCE: Dep. Anat. Cell Biol., McGill Univ., Montreal, QC, H3A 2B2, Can.
SOURCE: Biology of Reproduction (**1994**), 50(2), 309-19
CODEN: BIREBV; ISSN: 0006-3363
DOCUMENT TYPE: Journal
LANGUAGE: English
AB To determine whether male germ cells contain specific storage sites for poly(A)+ RNAs, in situ hybridizations were performed with sections of rat testis and a [3H]polyuridylic acid probe. The highest levels of poly(A)+ RNA were found in spermatocytes and round spermatids, while lower levels of poly(A)+ RNA detected in spermatogonia, elongated spermatids, Sertoli cells, myoid cells, fibroblasts, macrophages, and Leydig cells. No poly(A)+ RNA was detected in residual bodies of elongated spermatids. At stages IX-XI of the seminiferous cycle, the nuclei and cytoplasm of pachytene spermatocytes contained approx. equal amts. of poly(A)+ RNA, suggesting nuclear storage and/or a reduced processing rate of mRNA precursors at this stage of germ cell differentiation. To examine the distribution of poly(A)+ RNAs in subcellular components of testicular cells, electron microscope radioautog. was used. In germ cells and Sertoli cells, poly(A)+ RNA was often seen free in the cytoplasm or associated with the endoplasmic reticulum and was only occasionally found associated with mitochondria, lysosomes, lipid inclusions, and axonemes. As previously reported for the mRNAs of transition protein 1 and **protamine** 1, no compartmentalization of poly(A)+ RNAs was detected in the cytoplasm of round and elongated spermatids. No poly(A)+ RNA was detected in association with the radial body and in most sections, the chromatoid body did not contain any significant amts. of poly(A)+ RNA.

L6 ANSWER 32 OF 34 CA COPYRIGHT 2004 ACS on STN
ACCESSION NUMBER: 44:25534 CA
ORIGINAL REFERENCE NO.: 44:5021b-d
TITLE: Distribution of desoxyribonuclease in the developing embryo (Arbacia punctulata)
AUTHOR(S): Mazia, Daniel
CORPORATE SOURCE: Univ. of Missouri, Columbia
SOURCE: Journal of Cellular and Comparative Physiology (**1949**), 34, 17-32
CODEN: JCCPAY; ISSN: 0095-9898
DOCUMENT TYPE: Journal

LANGUAGE: Unavailable

AB Desoxyribonuclease activity (I), measured by the **protamine** precipitation method, is distributed through the cytoplasm of the unfertilized egg. There is no evidence of **nuclear localization** of any considerable portion of I. In unfertilized eggs I is greater than in adult mammalian tissues. The pH optimum is approx. 7. The concentration of I in Arbacia spermatozoa, per cell, is approx. 10-4 that of the egg, but per unit volume it is approx. 10 times that of the egg. Total I does not change during the 1st 40 hrs. of development but the sedimentable (20,000 g., 15 min.) fraction increases from an insignificant amount in unfertilized eggs to 80% of the total at 40 hrs. This change in the sedimentable fraction parallels the increase in desoxyribonucleic acid.

L6 ANSWER 33 OF 34 SCISEARCH COPYRIGHT 2004 THOMSON ISI on STN
ACCESSION NUMBER: 90:393640 SCISEARCH
THE GENUINE ARTICLE: DN794
TITLE: IMMUNOCYTOCHEMICAL **LOCALIZATION** OF **NUCLEAR PROTAMINE** IN BOAR SPERMATOZOA DURING EPIDIDYMAL TRANSIT
AUTHOR: RODRIGUEZMARTINEZ H (Reprint); COURTENS J L; KVIST U; PLOEN L
CORPORATE SOURCE: SWEDISH UNIV AGR SCI, FAC VET MED, DEPT ANAT & HISTOL, BOX 7011, S-75007 UPPSALA, SWEDEN (Reprint); KAROLINSKA HOSP, DEPT CLIN CHEM, ANDROL UNIT, STOCKHOLM, SWEDEN; INRA, PHYSIOL REPROD STN, F-37380 MONNAIE, FRANCE
COUNTRY OF AUTHOR: SWEDEN; FRANCE
SOURCE: JOURNAL OF REPRODUCTION & FERTILITY, (**1990**) Vol. 89, No. 2, pp. 591-595.
DOCUMENT TYPE: Article; Journal
FILE SEGMENT: LIFE
LANGUAGE: ENGLISH
REFERENCE COUNT: 27

L6 ANSWER 34 OF 34 SCISEARCH COPYRIGHT 2004 THOMSON ISI on STN
ACCESSION NUMBER: 88:327294 SCISEARCH
THE GENUINE ARTICLE: N7147
TITLE: IMMUNOCYTOCHEMICAL **LOCALIZATION** OF **NUCLEAR PROTAMINES** IN BOAR SPERM DURING EPIDIDYMAL TRANSIT
AUTHOR: RODRIGUEZMARTINEZ H (Reprint); JANSSON A; COURTENS J L; PLOEN L
CORPORATE SOURCE: SWEDISH UNIV AGR SCI, FAC VET MED, DEPT ANAT & HISTOL, S-75007 UPPSALA, SWEDEN; INRA, PHYSIOL REPROD STN, F-37380 MONNAIE, FRANCE
COUNTRY OF AUTHOR: SWEDEN; FRANCE
SOURCE: JOURNAL OF ULTRASTRUCTURE AND MOLECULAR STRUCTURE RESEARCH , (**1988**) Vol. 98, No. 3, pp. 327.
DOCUMENT TYPE: Conference; Journal
FILE SEGMENT: LIFE
LANGUAGE: ENGLISH
REFERENCE COUNT: 2

=> d his

(FILE 'HOME' ENTERED AT 13:59:28 ON 24 MAR 2004)

FILE 'MEDLINE, BIOSIS, EMBASE, CA, SCISEARCH' ENTERED AT 13:59:34 ON 24 MAR 2004
L1 30366 S PROTAMINE
L2 4 S (S (N) PROTAMIN? (N) NLS)
L3 1 DUP REM L2 (3 DUPLICATES REMOVED)

```
L4          107 S L1 AND (NUCLE? (2N) LOCAL?)
L5           47 DUP REM L4 (60 DUPLICATES REMOVED)
L6           34 S L5 AND PY=<2001

=> s l4 and (s (2n) protamin?)
L7            9 L4 AND (S (2N) PROTAMIN?)

=> dup rem l7
PROCESSING COMPLETED FOR L7
L8            2 DUP REM L7 (7 DUPLICATES REMOVED)

=> d l8 1-2 ibib abs

L8   ANSWER 1 OF 2     MEDLINE on STN                    DUPLICATE 1
ACCESSION NUMBER:   2002200709     MEDLINE
DOCUMENT NUMBER:    PubMed ID: 11906253
TITLE:              Protamine-fragment peptides fused to an SV40
                    nuclear localization signal deliver
                    oligonucleotides that produce antisense effects in prostate
                    and bladder carcinoma cells.
AUTHOR:             Benimetskaya Luba; Guzzo-Pernell Nancy; Liu Su-Ting; Lai
                    Johnathan C H; Miller Paul; Stein C A
CORPORATE SOURCE:   Howard Florey Institute of Experimental Physiology and
                    Medicine, University of Melbourne, Parkville, Victoria,
                    3052, Australia.
CONTRACT NUMBER:    R01 GM58791 (NIGMS)
SOURCE:             Bioconjugate chemistry, (2002 Mar-Apr) 13 (2) 177-87.
                    Journal code: 9010319. ISSN: 1043-1802.
PUB. COUNTRY:       United States
DOCUMENT TYPE:      Journal; Article; (JOURNAL ARTICLE)
LANGUAGE:           English
FILE SEGMENT:       Priority Journals
ENTRY MONTH:        200207
ENTRY DATE:         Entered STN: 20020406
                    Last Updated on STN: 20020724
                    Entered Medline: 20020723
AB  The development of antisense technology has focused on improving methods
    for oligonucleotide delivery into cells. In the present work, we describe
    a novel strategy for oligonucleotide delivery based on a bifunctional
    peptide composed of a C-terminal protamine-fragment that
    contains a DNA-binding domain and an N-terminal nuclear
    localization signal sequence derived from the SV40 large-T antigen
    (The sequences of two of the peptides are R6WGR6-PKKKRKV [s-
    protamine-NLS] and R4SR6FGR6VWR4-PKKKRKV [l-protamine
    -NLS]). We demonstrated, by intrinsic fluorescence quenching, that
    peptides of this class form complexes with oligodeoxynucleotides. To
    evaluate delivery, we used a 20-mer phosphorothioate oligomer (Isis 3521)
    targeted to the 3'-untranslated region of the PKC-alpha mRNA and G3139, an
    18-mer phosphorothioate targeted to the first six codons of the human
    bcl-2 open reading frame, and complexed them with either of two peptides (
    s- or l-protamine-NLS). These peptides bind to and
    deliver antisense oligonucleotides to the nucleus of T24 bladder and PC3
    prostate cancer cells, as demonstrated by confocal microscopy.
    Furthermore, as shown by Western and Northern blotting, the
    peptide-oligonucleotide complexes produced excellent downregulation of the
    expression of the complementary mRNAs, which in turn resulted in
    downregulation of protein expression. However, under certain
    circumstances (predominantly in PC3 cells), incubation of the cells with
    chloroquine was required to produce antisense activity. Using this
    strategy, PKC-alpha protein and mRNA expression in T24 and PC3 cells and
    bcl-2 expression in PC3 cells was reduced by approximately 75 +/- 10% at a
    minimum concentration of oligomer of 0.25 microM, in combination with
    12-15 microM peptide. On the basis of our results, we conclude that
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arginine-rich peptides of this class may be potentially useful delivery vehicles for the cellular delivery of antisense oligonucleotides. This new strategy may have several advantages over other methods of oligonucleotide delivery and may complement already existing lipid-based technologies.

L8 ANSWER 2 OF 2 MEDLINE on STN DUPLICATE 2
ACCESSION NUMBER: 1998010146 MEDLINE
DOCUMENT NUMBER: PubMed ID: 9349433
TITLE: **Protamine** sulfate enhances lipid-mediated gene transfer.
AUTHOR: Sorgi F L; Bhattacharya S; Huang L
CORPORATE SOURCE: Department of Pharmacology, University of Pittsburgh School of Medicine, PA 15261, USA.
CONTRACT NUMBER: CA 59327 (NCI)
CA 64654 (NCI)
CA 71731 (NCI)
+
SOURCE: Gene therapy, (1997 Sep) 4 (9) 961-8.
Journal code: 9421525. ISSN: 0969-7128.
PUB. COUNTRY: ENGLAND: United Kingdom
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 199711
ENTRY DATE: Entered STN: 19971224
Last Updated on STN: 19971224
Entered Medline: 19971120

AB A polycationic peptide, **protamine** sulfate, USP, has been shown to be able to condense plasmid DNA efficiently for delivery into several different types of cells in vitro by several different types of cationic liposomes. The monovalent cationic liposomal formulations (DC-Chol and lipofectin) exhibited increased transfection activities comparable to that seen with the multivalent cationic liposome formulation, lipofectamine. This suggests that lipofectamine's superior in vitro activity arises from its ability to condense DNA efficiently and that **protamine**' **s** primary role is that of a condensation agent, although it also possesses several amino acid sequences resembling that of a **nuclear localization** signal. While the use of polycations to condense DNA has been previously reported, the of **protamine** sulfate, USP as a condensation agent was found to be superior to poly-L-lysine as well as to various other types of **protamine**. These differences among various salt forms of **protamine** appear to be attributable to structural differences between the **protamines** and not due to differences in the net charge of the molecule. The appearance of lysine residues within the **protamine** molecule correlate with a reduction in binding affinity to plasmid DNA as well as an observed loss in transfection enhancing activity. This finding sheds light on the structural requirements of condensation agents for use in gene transfer protocols. Furthermore, **protamine** sulfate, USP is an FDA-approved compound with a documented safety profile and could be readily used as an adjuvant to a human gene therapy protocol.